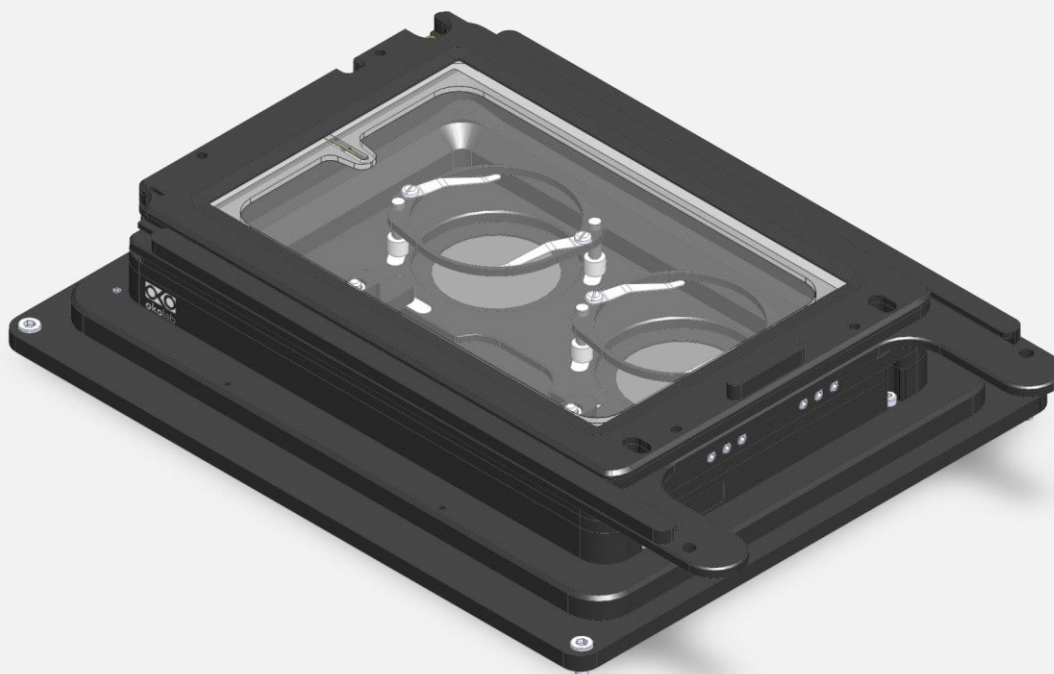




H301-OLYMPUS-IX3-SVR

Compatible with the following XY stage

- Olympus IX3-SVR

Compatible with the following Okolab Controllers

- H301-T-BL-PLUS
- UNO-COMBINED-CONTROLLER
- H401-T-DUAL

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1. Components and dimensions

H301-OLYMPUS-IX3-SVR includes the following components:

- **Chamber main body** with embedded temperature sensor. It connects on the stage with screws.
- **Sliding heated glass lid** with Indium Tin Oxide (ITO) conductive coating and embedded temperature sensor.
- **Chamber riser**. It is a removable frame increasing the height of the chamber from 27 to 33 mm, often required when using multi-well (MW) plates.

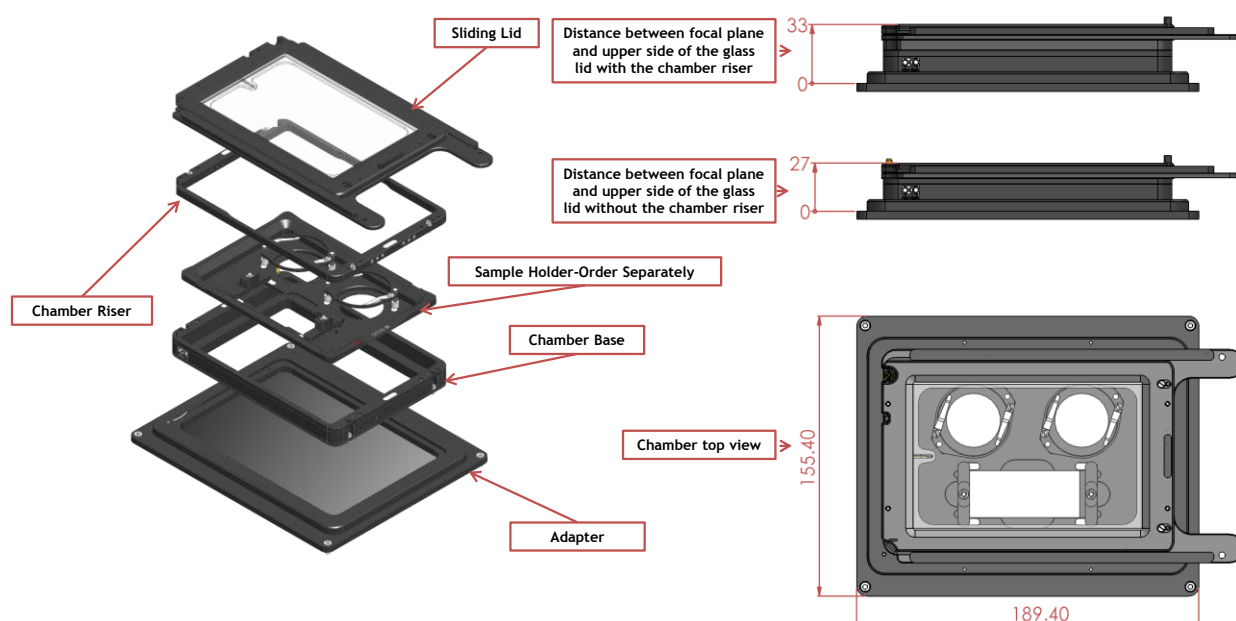


Figure 1. H301-OLYMPUS-IX3-SVR - Components and Dimensions.

2. Sample Holders

2.1 Available Sample Holders

The following sample holders are available.

NOTE: Please contact info@oko-lab.com if you cannot find the sample holder you are looking for. We are constantly adding new inserts to the list.

1x35-M	#1 35mm Petri-dish
1xGS-M	#1 1x3in. chamber slide
1xLABTEK-M	#1 Lab-Tek 1in.x2in. chambered cover glass
1xLABTEK-II-M	#1 Lab-Tek II 1in.x2in. chambered cover glass
1x60-M	#1 60mm Petri-dish
1xT25-M	#1 Nunc and Greiner T25 flask
2x35-M	#2 35mm Petri-dish
2xGS-M	#2 1x3in. chamber slides
2xLABTEK-M	#2 Lab-Tek 1in.x2in. chambered cover glass

2xLABTEK-II-M	#2 Lab-Tek II 1in.x2in. chambered cover glass
2x60-M	#2 60mm Petri-dish
4x35-M	#4 35mm Petri-dish
GS35-M	#1 1x3in. chamber slide and #2 35mm Petri-dish
LABTEK-35-M	#1 Lab-Tek 1in.x2in. chambered cover glass and #2 35mm Petri-dish
LABTEK-II-35-M	#1 Lab-Tek II 1in.x2in. chambered cover glass and #2 35mm Petri-dish
LABTEK-60-M	#1 Lab-Tek 1in.x2in. chambered cover glass and #1 60mm Petri-dish
LABTEK-II-60-M	#1 Lab-Tek II 1in.x2in. chambered cover glass and #1 60mm Petri-dish
GS60-M	#1 1x3in. chamber slide and #1 60mm Petri-dish
6035-M	#1 60mm Petri-dish and #1 35mm Petri-dish
6MW	6-well plates, one MW LOCK required
12MW	12-well plates, one MW LOCK required
24MW	24-well plates, one MW LOCK required
24MW-NUNC	24-well NUNC/GREINER plates, one MW LOCK required
48MW	48-well plates, one MW LOCK required
96MW	96-well plates, one MW LOCK required. It includes the insert for oil immersion imaging.
MW-LOCK-22.5	Magnetic lock to hold the standard MW plates in the chamber
MW-LOCK-16.5	Magnetic lock to hold the low-profile MW plates in the chamber



Figure 2. Available sample holders.

3. Available Lids

The following heated glass Lids are available for H301-OLYMPUS-IX3-SVR:

- **H301-SLIDING-LID:** For easy sample loading and pipetting - **Included in code H301-OLYMPUS-IX3-SVR**
- **H301-KOEHLER-LID*:** Reduces chamber height to 22 mm and allows imaging under Koehler illumination
- **H301-INJECTION-LID*:** Heated glass lid with two small openings (sealed with flexible plastic) allowing injection or permanent access to the sample. Compatible only with sample holder 1x35
- **H301-LASER-INTERLOCK-LID*:** Heated glass lid with safety switch. Connects to laser controller and automatically turns laser off when lid is lifted. Compatible only with sample holder 1x35

* OPTIONAL - not included with H301-OLYMPUS-IX3-SVR

3.1 Sliding lid

The Sliding Lid is a heated glass lid allowing for easy sample loading and pipetting. The Sliding Lid is screwed onto the chamber.

Figure 3 shows chamber dimensions with the Sliding Lid (with and without chamber riser). Figure 4 illustrates how to remove Sliding Lid, when a different Lid is necessary.

NOTE: Sliding Lid MUST BE REMOVED when using any other Lid. The Sliding Lid is fixed onto the chamber with 4 screws. Screws location is indicated by letter A in Figure 4 (image 2 and 3). Keep Sliding Lid fully closed to access screws labeled A in image 2. Open Sliding Lid to access screws labeled A in image 3.

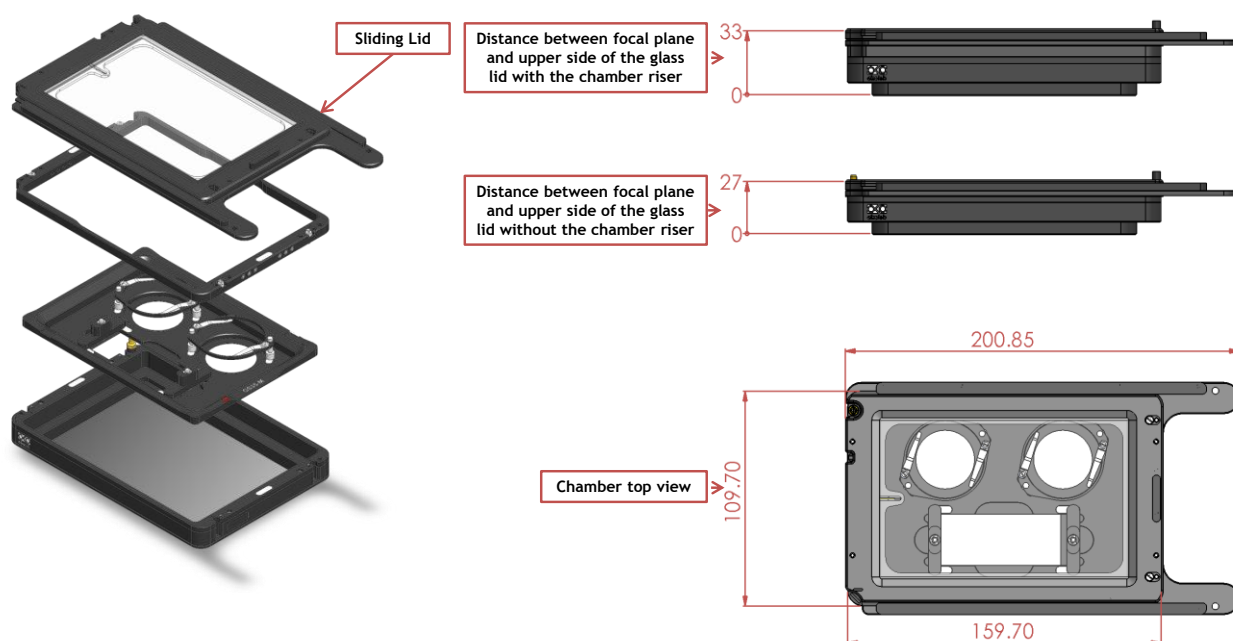


Figure 3. H301-OLYMPUS-IX3-SVR (Sliding Lid comes as standard)

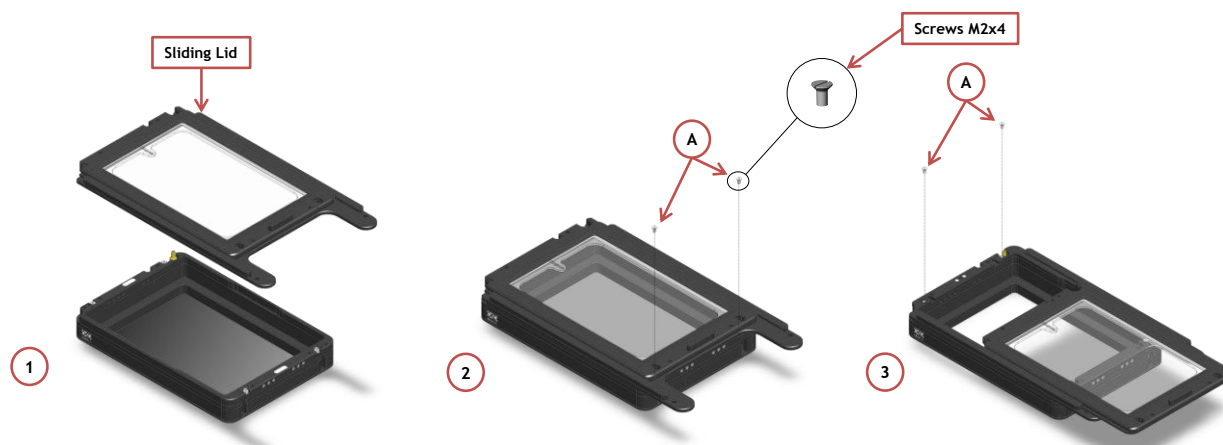


Figure 4. Assembly of the sliding lid

3.2 Koehler Lid

The Koehler Lid is a heated glass lid reducing the chamber's height to 22 mm and allowing imaging under Koehler illumination. The Koehler Lid is placed onto the chamber (NO SCREWS or TOOLS are necessary).

Figure 5 shows chamber dimensions with Koehler lid (with and without chamber riser).

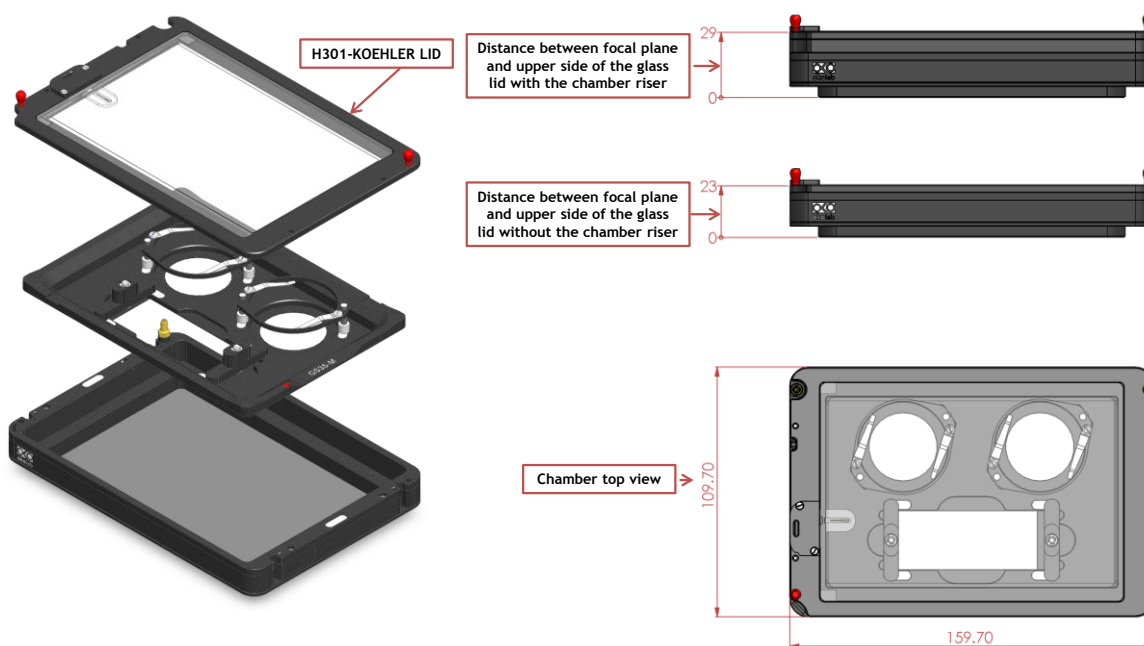


Figure 5. H301-OLYMPUS-IX3-SVR + H301-KOEHLER-LID

3.3 Injection Lid

The Injection Lid is a heated glass lid with two small openings sealed with flexible plastic allowing injection or permanent access to the sample. NOTE: Compatible with sample holder 1x35 ONLY. The Injection Lid is placed onto the chamber (NO SCREWS or TOOLS are necessary).

Figure 6 shows chamber dimensions with the Injection Lid (with and without chamber riser).

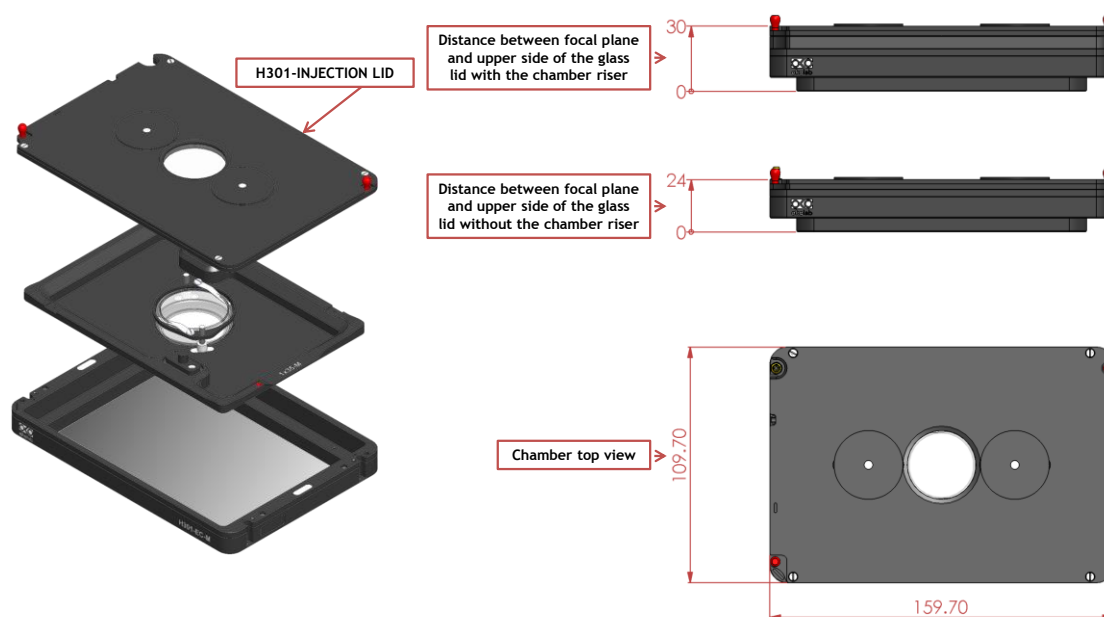


Figure 6. H301-OLYMPUS-IX3-SVR+ H301-INJECTION-LID

3.4 Laser Interlock Lid

The Laser Interlock Lid is a heated glass lid with a safety switch connecting to the laser controller. It automatically turns the laser off when the lid is lifted. NOTE: Compatible with sample holder 1x35 ONLY. The Laser Interlock Lid is placed onto the chamber (NO SCREWS or TOOLS are necessary).

Figure 7 shows chamber dimensions with the Laser Interlock Lid (with and without chamber riser).

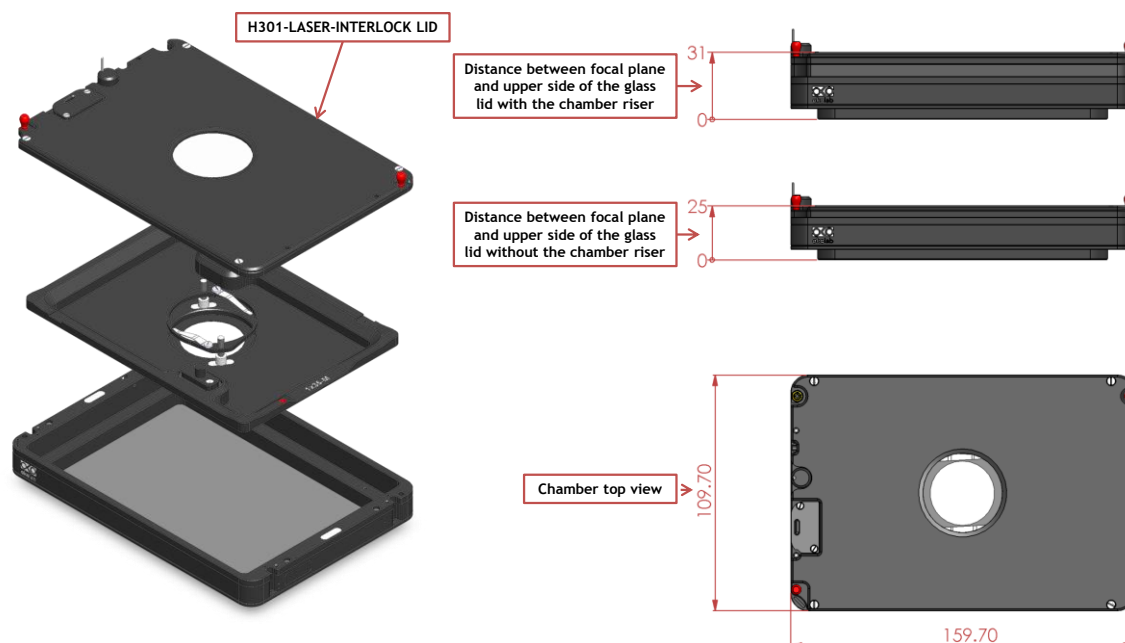


Figure 7. H301-OLYMPUS-IX3-SVR + H301-LASER-INTERLOCK-LID

4. Insertion of the Sample Feedback Temperature Sensor

Insert the Sample Feedback Temperature Sensor through the dedicated opening located in the H301-OLYMPUS-IX3-SVR (see Figure 8, Front and 3D views).

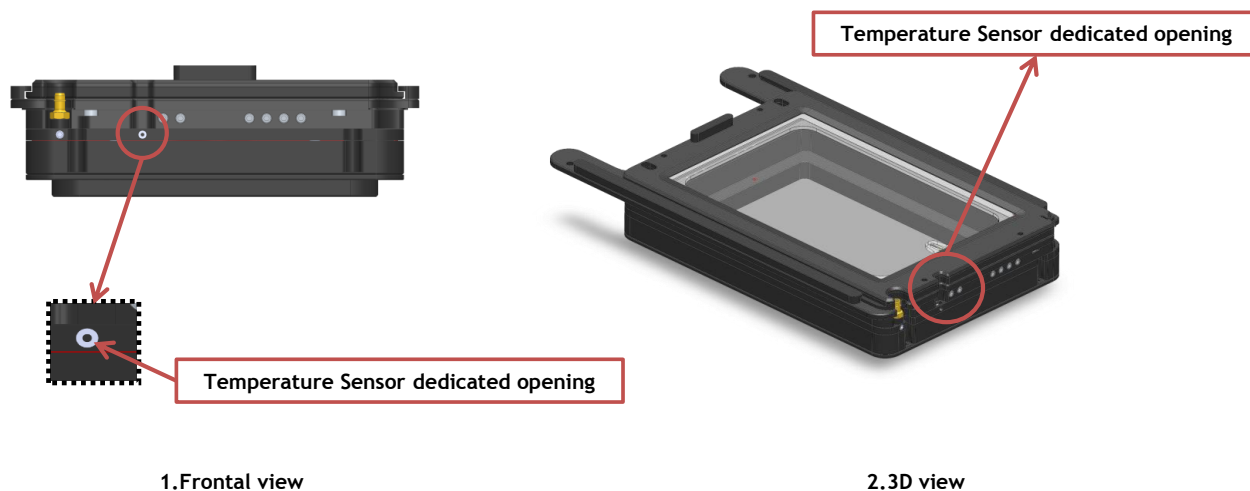


Figure 8. Insertion of the temperature sensor inside the chamber.

5. Insertion of Sample Holder into Chamber

Sample holders fit into the chamber base and are held in place by magnets embedded within both chamber and holder. To introduce a sample holder with the proper orientation, match the red dot on the holder to the one on the chamber base, as illustrated in Figure 9.

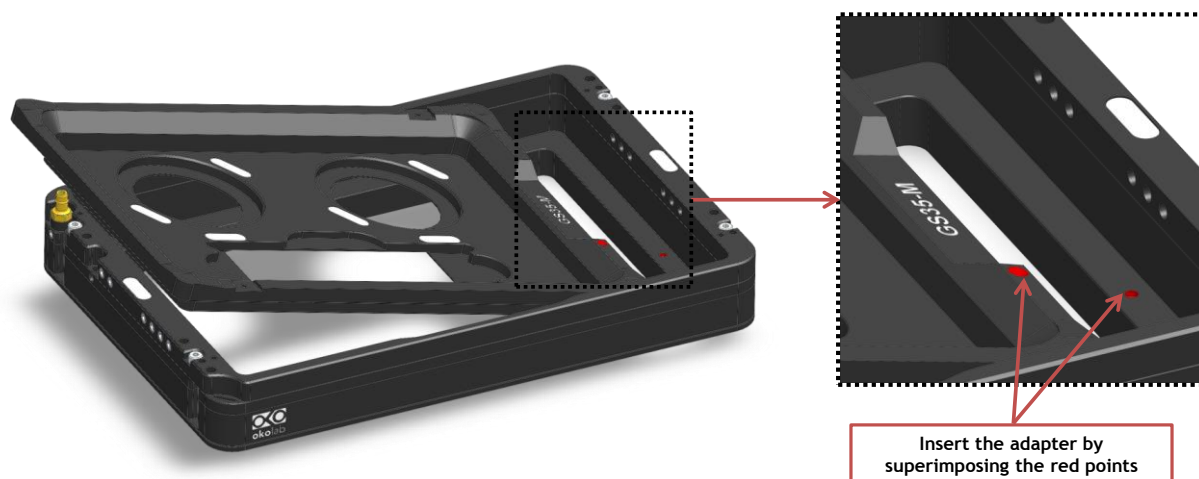


Figure 9. Introduction of the Specimen Holder inside the Chamber Base.

6. Working with 35 or 60 mm Petri Dish – Spacing Rings and Magnetic Locks

Magnetic locks prevent movement of 35 and 60 mm dishes inside the sample holder.

Figure 10 illustrates the available magnetic locks for 35 and 60 mm dishes. Threaded magnetic posts allow adjusting holder's height.

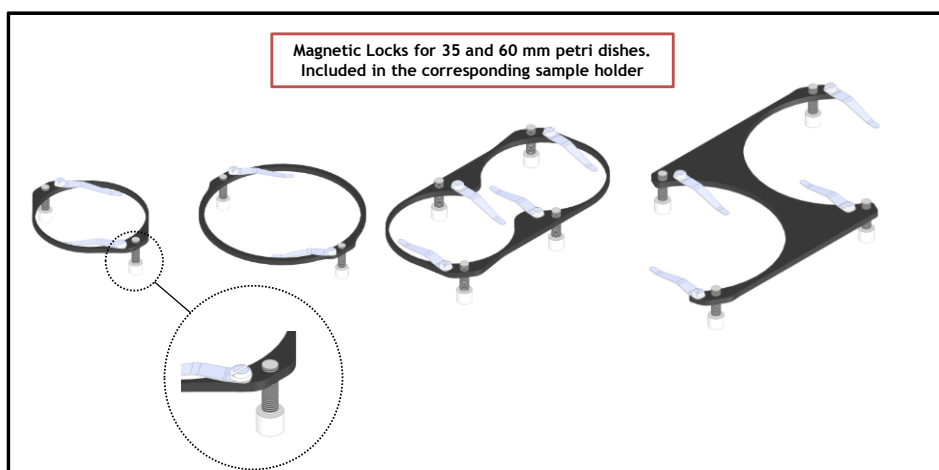


Figure 10. Magnetic locks for 35 and 60 mm dish

NOTE: Magnetic locks are included with sample holder.

Spacing rings accounting for variability in dish diameter from one manufacturer to another are also included. Figure 11 shows how to position the ring. Ring selection guide diagrams: rings for 35 and 60 mm dish -Figure 12 and Figure 13, respectively.

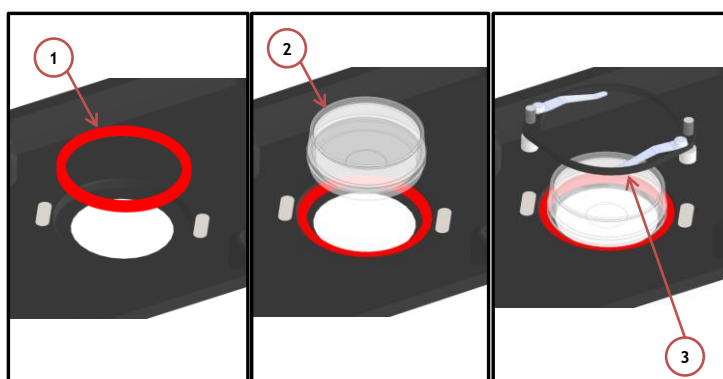


Figure 11. 1) Insertion of the ring, 2) insertion of the 35 mm dish, 3) insertion of the magnetic lock.

NOTE: Spacing rings are included with sample holder.

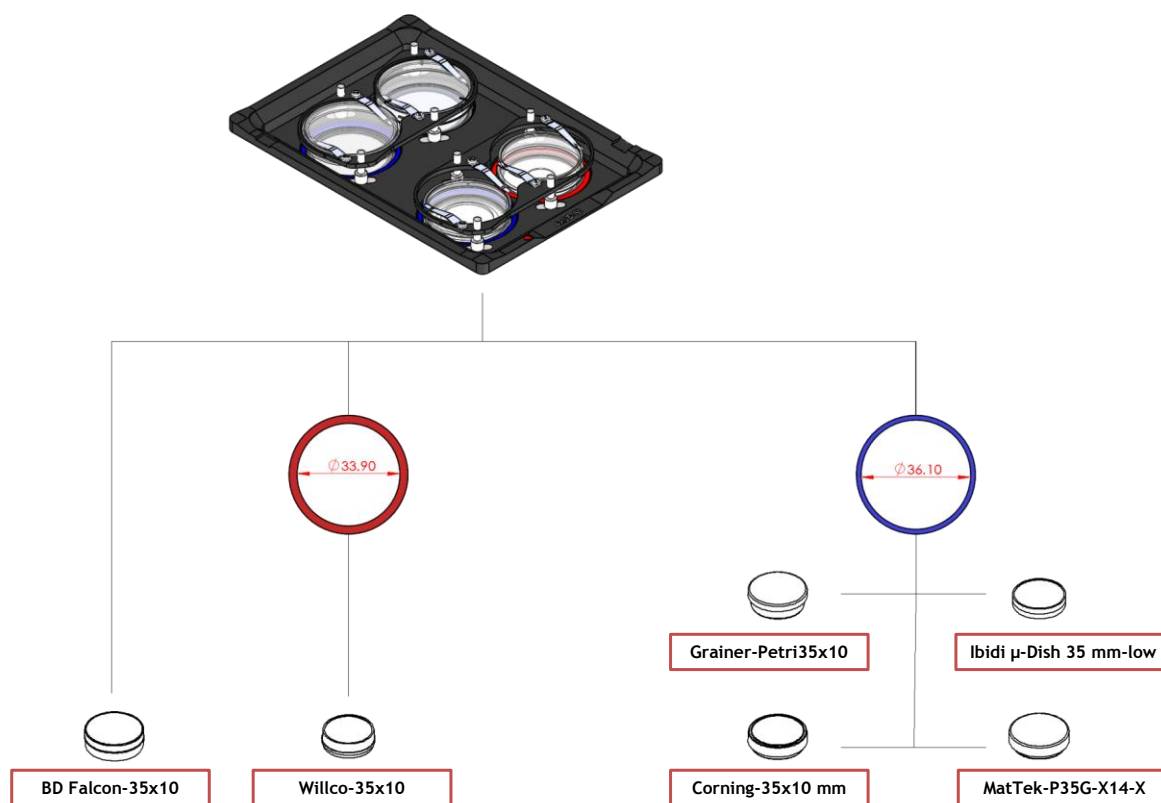


Figure 12. Ring selection scheme for 35 mm dish.

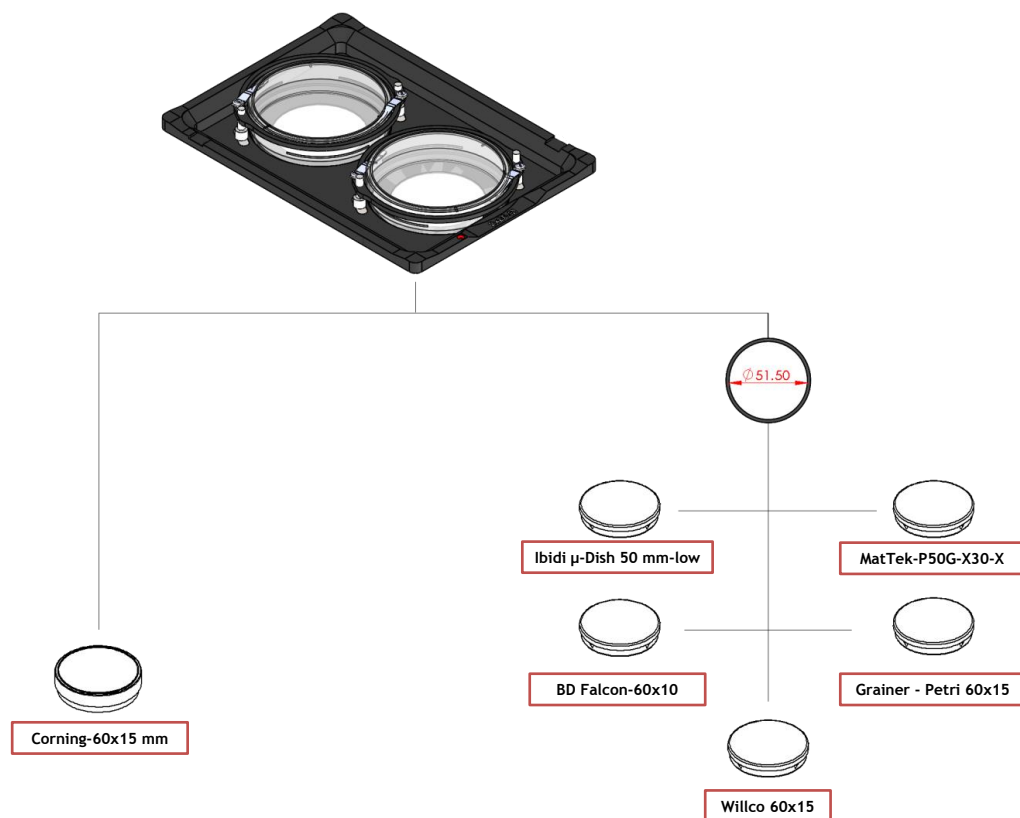


Figure 13. Ring selection scheme for 60 mm dish.

7. Working with 1x3'' and 1x2'' chamber slides - magnetic locks

Magnetic locks prevent movement of 1''x 3'' and 1''x 2'' chamber slides inside of the sample holder.

NOTE: Magnetic locks are included with sample holder.

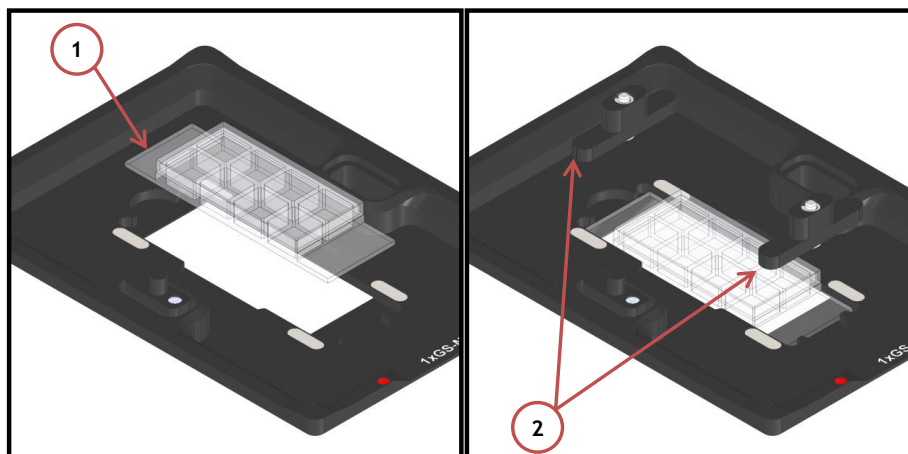


Figure 14. Magnetic lock for 1'' x 3'' chamber slide.

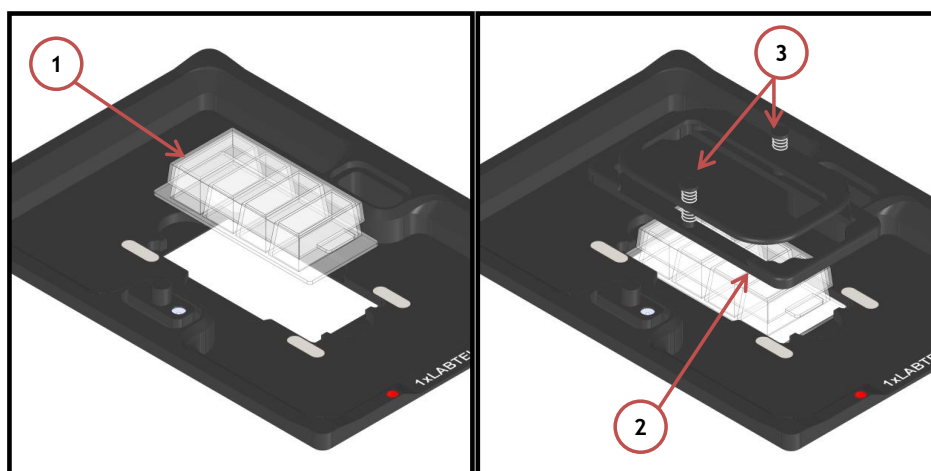


Figure 15. Magnetic lock for 1'' x 2'' chamber slide. To lock the slide, push simultaneously the buttons indicated with (3).

8. Working with MW Plates - Magnetic locks and Chamber riser

Magnetic locks hold MW plates in place into sample holder illustrated in Figure 16

NOTE: Magnetic locks must be ordered separately (NOT INCLUDED). Product codes: MW-LOCK 22.5 and MW-LOCK 16.5.

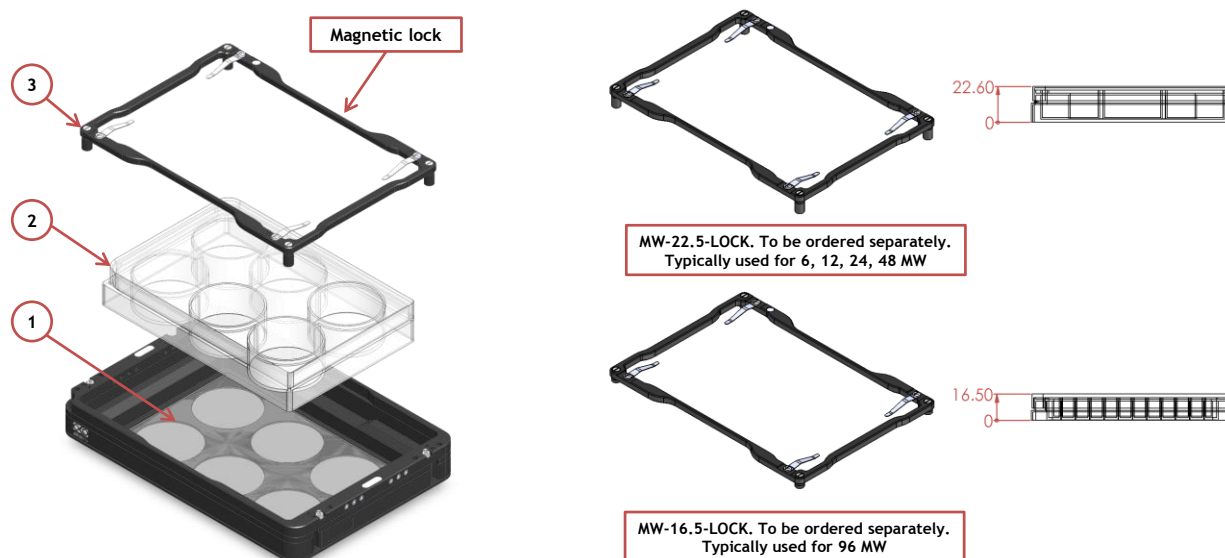


Figure 16. Magnetic locks for MW plates. To be ordered separately.

Chamber Riser increases chamber's height and is REQUIRED with 6, 12, 24, 48 MW plates, regardless of the Lid employed.

Figure 17 shows how to install and fasten the chamber riser. Screws location is indicated by letter A.

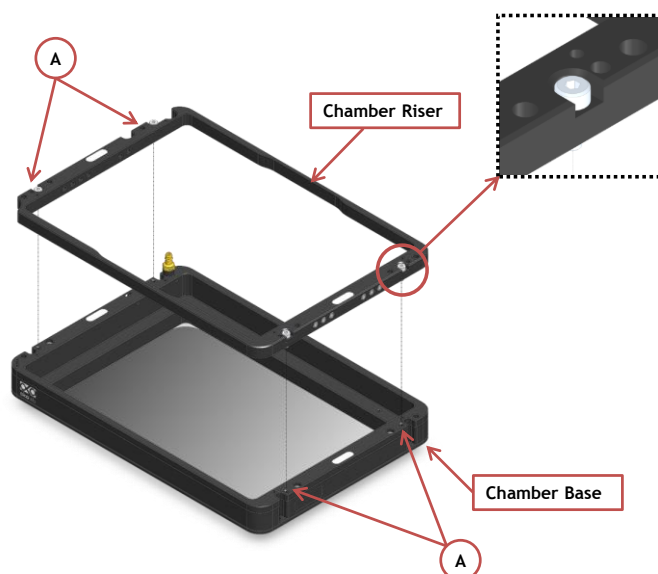


Figure 17. Riser Assembly.

9. Connection of the Gas Supply

A single silicon tubing carries output gas from the Okolab Gas Controller to the H301-OLYMPUS-IX3-SVR. Silicon tubing connects to a gas input - brass opening - located on a corner of the H301-OLYMPUS-IX3-SVR. See Figure 18. Connect by gently pushing silicon tubing onto brass opening.



Figure 18. Connection with gas supply.

10. Working with Perfusion

The Chamber Riser included with H301-OLYMPUS-IX3-SVR features 12 perfusion holes for the insertion of perfusion tubing up to 2.5 mm in outer diameter. Small screws plug the perfusion holes when not in use. (Grub screws M3x6). Remove small screws as necessary before introducing perfusion tubing.

Figure 19 shows location of perfusion holes.

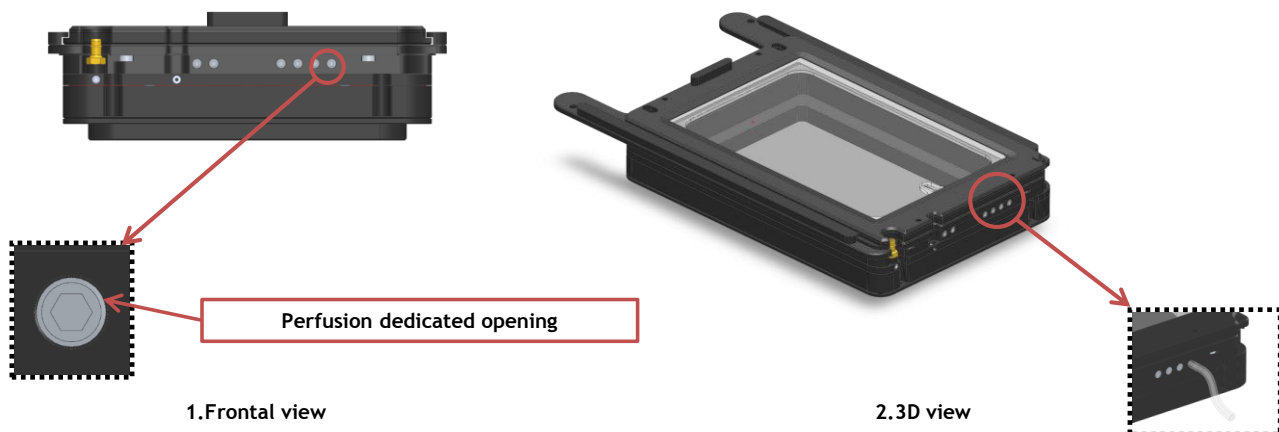


Figure 19. Perfusion

11. Connecting the Chamber to the adapter

Follow the steps shown in the images of Figure 20 and listed below in order to correctly connect the chamber with its adapter:

1. Follow the arrows shown in Figure 20 Images 1 and 2 for placing the captive screws in the proper housings
2. Place the chamber on the adapter as shown in, Figure 20 Image 3
3. Use a 1.5mm metric Allen Wrench to tighten two of the four captive screws in place (A in Image 4) while keeping the sliding lid closed. (See Figure 20 Image 4)

4. Tighten the remaining two captive screws (A in Image 5) while keeping the chamber sliding lid open.
5. Image 6 of Figure 20 shows the chamber fixed on the adapter

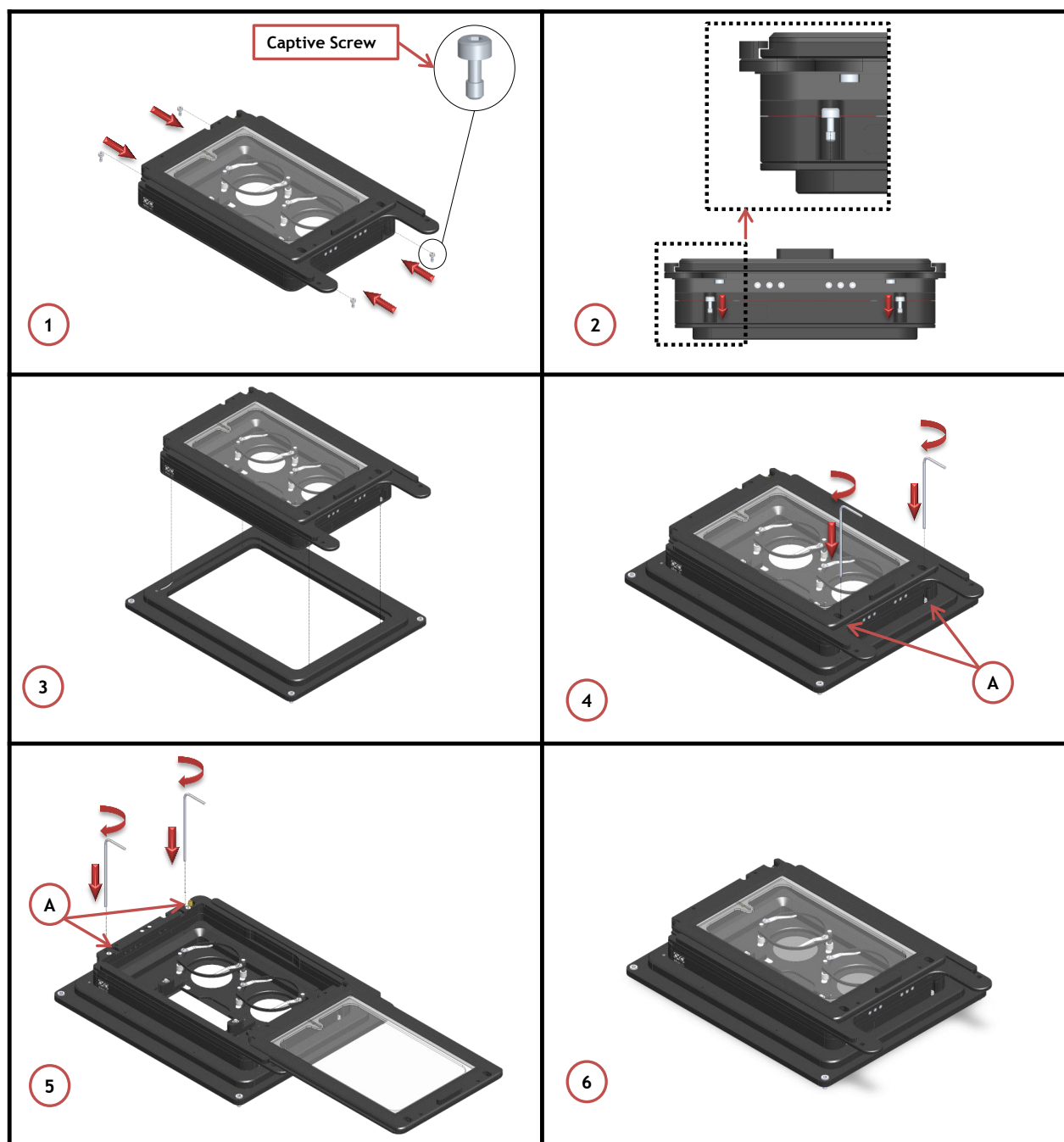


Figure 20. How to assemble the chamber to the adapter.

12. Connection of the Chamber with XY stage

Once you have assembled the chamber with adapter you can place it on the XY stage, following the steps shown in Figure 21.

1. Place the adapter on the stage (See Image 1 of Figure 21).
2. Follow the instructions shown in Image 2 of Figure 21 for placing socket head cap screws; use the provided Allen wrench for tightening the socket head cap screws and fastening the adapter on the stage. Screws housings are indicated with letter A in Image 2 of Figure 21.

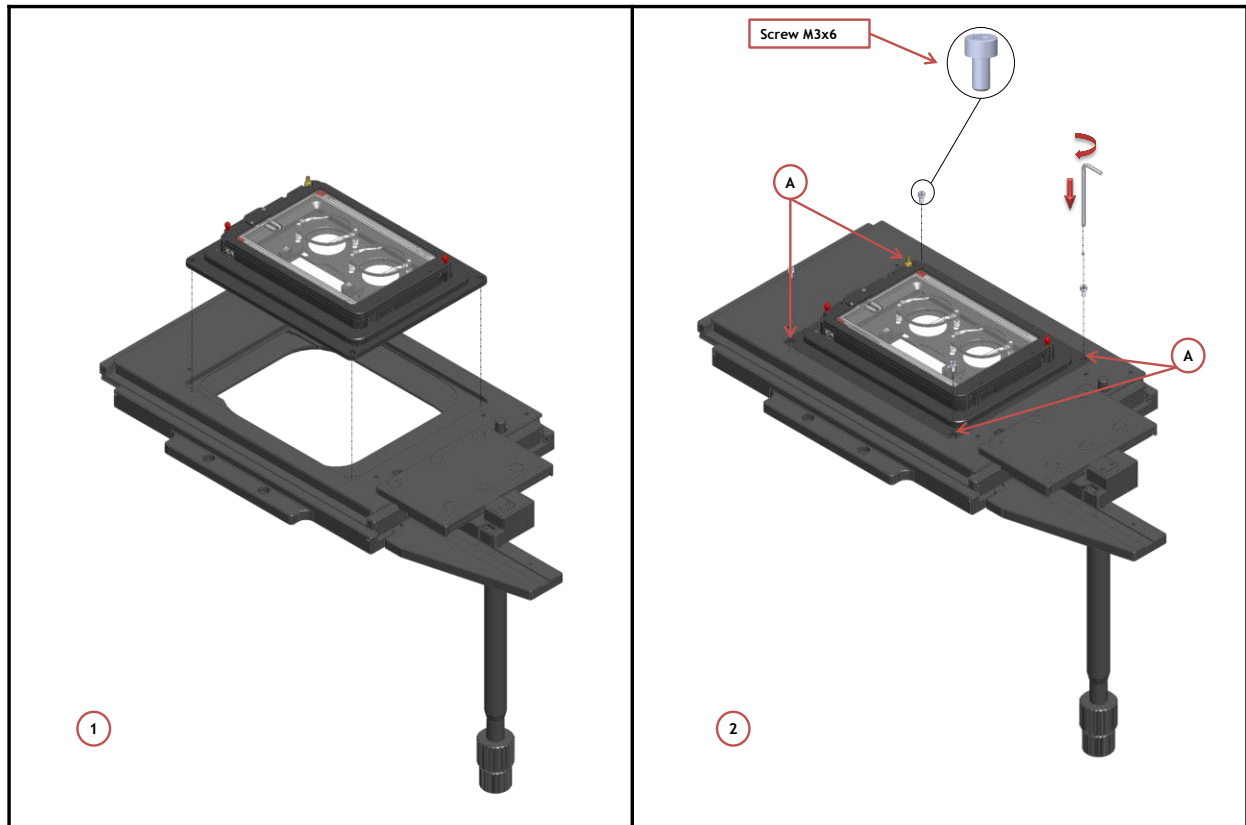


Figure 21. How to fix the H301-OLYMPUS-IX3-SVR to the XY stage.